

Potential for Sexual Reproduction of *Phytophthora ramorum* in Washington State Nurseries¹

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Abstract

In 2003, isolates belonging to the A1 mating type were reported from commercial nurseries in Oregon. Soon thereafter, we reported the presence of both mating types of *P. ramorum* in nurseries in Washington. AFLP, microsatellite, and RFLP of the Cox I region indicated the A1 isolates belonged to the European (E.U.) lineage of *P. ramorum*. In 2003, 33 percent of the isolates analyzed belonged to European lineage, while the rest belonged to the North American (N.A.) lineage. Isolates belonging to the two lineages were found in the same block of plants, and even on adjacent plants. By using tester strains (A2 from California and A1 from Oregon nurseries) we confirmed that all North American isolates belonged to the A2 mating type, and all of the European isolates belonged to the A1 mating type. The coexistence of both mating types on adjacent plants increases the chances for sexual recombination between these two genetically divergent lineages. Crosses with tester strains were 100 percent successful, but a very high proportion of oospores appeared to be aborted, suggesting the mating system of *P. ramorum* is not perfectly functional, potentially due to the long isolation of the two mating types. Nonetheless, a significant number of oospores appeared to be viable. In 2004, the survey of isolates from nurseries in Washington continued. PCR-RFLP of the Cox I region and morphology of the colony (appressed for N.A. and fluffy for E.U. isolates) indicated that 84 appressed colonies had the E.U. RFLP pattern, while only one isolate had fluffy colony morphology and had a RFLP pattern matching that of the E.U. lineage. Despite the sharp decrease of A1 isolates in 2004, we encountered one isolate that had fluffy colony morphology and RFLP pattern of the N.A. isolates. Out of 12 microsatellite loci analyzed for this isolate, two had alleles exclusively found in North America, two had alleles exclusively found in Europe, one had one allele that is shared by both lineages, and seven loci had novel alleles. AFLP analysis showed this isolate has bands typical of the N.A. lineage, bands typical of the E.U. lineage, and a significant number of new bands. This isolate is likely to be either a recombinant between A1 and A2 or an isolate representative of a third yet unknown lineage. Mating and phenotype of this isolate were discussed, including implications for the evolution of *P. ramorum*.

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